

## CLAIMS

1. A method of disinfecting article(s) that are susceptible to contamination by infectious prion protein, the method comprising the steps of:
  - (a) heating said article(s) to a sufficient temperature and for sufficient time to enhance the proteolytic susceptibility of infective prion protein associated with said article(s); and
  - (b) exposing the heated article(s) to a proteolytic enzyme that is effective for at least partial reduction of the infective protein prion associated with said article(s).
2. The method of claim 1, wherein said articles comprise surgical instruments.
3. The method of claim 2, wherein said surgical instrument(s) are selected from the group consisting of: clamps, forceps, scissors, knives, cables, punches, tweezers, cannulae, calipers, carvers, curettes, scalers, dilators, clip applicators, retractors, contractors, excavators, needle holders, suction tubes, trocars, coagulation electrodes, electroencephalographic depth electrodes, rib and sternum spreaders, bipolar probes, and rib shears.
4. The method of claim 1, wherein said article(s) comprise cutleries and kitchen utensils.
5. The method of claim 4, wherein said cutleries and kitchen utensils are selected from the group consisting of: knives, forks, scissors, peelers, parers, slicers, spatulas, and cleavers.
6. The method of claim 1, wherein said article(s) comprise laboratory apparatus(es).

7. The method of claim 6, wherein said laboratory apparatus(es) are selected from the group consisting of: containers, filtration devices, centrifuges, spectrophotometers, and fluorometers.
8. The method of claim 1, wherein said article(s) comprise veterinary devices.
9. The method of claim 8, wherein said veterinary devices are selected from the group consisting of clamps, forceps, knives, saws, probes, and electronic stun equipment.
10. The method of claim 1, wherein the temperature in step (a) comprises a temperature not exceeding about 150°C.
11. The method of claim 1, wherein the temperature in step (a) comprises a temperature of at least 35°C.
12. The method of claim 1, wherein the temperature in step (a) comprises a temperature below about 150°C.
13. The method of claim 1, wherein the temperature in step (a) comprises a temperature in a range of from about 100°C to about 150°C.
14. The method of claim 1, wherein the temperature in step (a) comprises a temperature in a range of from about 125°C to about 140°C.
15. The method of claim 1, wherein step (b) is conducted at lower temperature than step (a).
16. The method of claim 1, wherein step (b) is carried out at temperature above about 40°C.
17. The method of claim 1, wherein step (b) is carried out at temperature above about 50°C.

18. The method of claim 1, wherein step (b) is carried out at temperature in a range of from about 35°C to about 75°C.
19. The method of claim 1, wherein step (b) is carried out at temperature in a range of from about 40°C to about 75°C.
20. The method of claim 1, wherein step (b) is carried out at temperature in a range of from about 50°C to about 65°C.
21. The method of claim 1, wherein the proteolytic enzyme comprises at least one enzyme selected from the group consisting of keratinase enzymes, proteinase K, trypsins, chymotrypsins, pepsins, chymosins, cathepsins, subtilisins, elastases, collagenases, endopeptidases, peptidases, oligopeptidase, thermolysins, bacillolysin, mycilysins, carboxypeptidases, leucyl aminopeptidases, aminopeptidases, extremthermophilic proteases, carbonyl hydrolase, papain, pancreatin, streptokinase, streptodornase, ficin, carboxypeptidase, chymopapain, and bromelin.
22. The method of claim 1, wherein the proteolytic enzyme comprises a keratinase enzyme.
23. The method of claim 1, wherein the proteolytic enzyme comprises an active fragment of a keratinase enzyme.
24. The method of claim 1, wherein the proteolytic enzyme comprises a *Bacillus licheniformis* PWD-1 enzyme or an active fragment thereof.
25. The method of claim 1, wherein the proteolytic enzyme comprises a protease enzyme.
26. The method of claim 25, wherein the protease enzyme comprises a carbonyl hydrolase.

27. The method of claim 26, wherein the carbonyl hydrolase comprises subtilisin.
28. The method of claim 27, wherein the subtilisin comprises a mutant of wild-type *Bacillus amyloliquefaciens* subtilisin, comprising one or more amino acid substitutions, additions, or deletions.
29. The method of claim 25, wherein the protease enzyme comprises at least one enzyme selected from the group consisting of: papain, pancreatin, trypsin, chymotrypsin, pepsin, streptokinase, streptodornase, ficin, carboxypeptidase, aminopeptidase, chymopapain, bromelin, and subtilisin.
30. A method of removing infective prion protein from a surgical instrument contaminated with same, the method including (a) heating the surgical instrument at a temperature in a range of from about 100°C to about 150°C, followed by (b) exposing the heated surgical instrument to a proteolytic enzyme at a temperature in a range of from about 35°C to about 100°C at which the proteolytic enzyme is thermally stable and proteolytically effective to at least partially destroy the infective prion protein contaminating said surgical instrument.
31. The method of claim 30, wherein said heating is conducted for a time of from about 5 minutes to about 5 hours.
32. The method of claim 30, wherein the proteolytic enzyme comprises at least one enzyme selected from the group consisting of keratinase enzymes, proteinase K, trypsins, chymotrypsins, pepsins, chymosins, cathepsins, subtilisins, elastases, collagenases, endopeptidases, peptidases, oligopeptidase, thermolysins, bacillolysin, mycilysins,

carboxypeptidases, leucyl aminopeptidases, aminopeptidases, extremothermophilic proteases, carbonyl hydrolase, papain, pancreatin, streptokinase, streptodornase, ficin, carboxypeptidase, chymopapain, and bromelin.

33. The method of claim 30, wherein the proteolytic enzyme comprises *Bacillus licheniformis* PWD-1 keratinase.
34. The method of claim 1, wherein the proteolytic enzyme comprises a protease enzyme.
35. The method of claim 34, wherein the protease enzyme comprises a carbonyl hydrolase.
36. The method of claim 35, wherein the carbonyl hydrolase comprises subtilisin.
37. The method of claim 36, wherein the subtilisin comprises a mutant of wild-type *Bacillus amyloliquefaciens* subtilisin, comprising one or more amino acid substitutions, additions, or deletions.
38. The method of claim 34, wherein the protease enzyme comprises at least one enzyme selected from the group consisting of: papain, pancreatin, trypsin, chymotrypsin, pepsin, streptokinase, streptodornase, ficin, carboxypeptidase, aminopeptidase, chymopapain, bromelin, and subtilisin.
39. A cleansing composition for disinfecting articles that are susceptible to contamination by infectious prion protein, said composition comprising:
  - (i) one or more proteolytic protein(s) selected from the group consisting of keratinase enzymes, proteinase K, trypsins, chymotrypsins, pepsins, chymosins, cathepsins, subtilisins, elastases, collagenases, endopeptidases, peptidases, oligopeptidase,

thermolysins, bacillolysin, mycilsins, carboxypeptidases, leucyl aminopeptidases, aminopeptidases, extremthermophilic proteases, carbonyl hydrolase, papain, pancreatin, streptokinase, streptodornase, ficin, carboxypeptidase, chymopapain, and bromelin; and

(ii) a solvent.

40. The cleansing composition of claim 39, comprising keratinase enzymes.
41. The cleansing composition of claim 40, wherein the concentration of said keratinase enzymes is with the range of from about 0.2 g/L to about 1.0 g/L.
42. The cleansing composition of claim 39, wherein the solvent is selected from the group consisting of distilled water, alcohol, buffer solution, and detergent solution.
43. The cleansing composition of claim 39, further comprising one or more chemical additives selected from the group consisting of surfactants, builders, boosters, and fillers.